201-16274

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June 2, 2006

Stephen L. Johnson, Administrator
U.S. Environmental Protection Agency
P.O. Box 1473
Merritield, VA 22 16

Attn: Chemical Right-to-Know Program

Re: EPA comments on the Test Plan and Robust Data Summary for Aminoalkylnitriles Category

Dear Administrator Johnson,

E. I. du Pont de Nemours & Company, Inc. received EPA's comments on the test plan and robust data summary for the aminoalkylnitriles category, which included propanenitile, 2-amino-2-methyl- (CAS#19355-69-2) and butanenitrile, and is pleased to respond. We have considered the recommended revisions to physiochemical properties, environmental fate, reproductive toxicity, developmental toxicity, repeated dose toxicity, genetic toxicity, and ecological sections. We have revised our submittal as needed on the attached summary sheet. Also included with this submittal is a revised robust data summary.

Please feel free to contact me with any questions or concerns you may have with regards to this submission at Edwin.L.Mongan-1@usa.dupont.com or by phone at 302-773-0910.

Sincerely,

Edwin L. Mongan, III
Manager, Environmental Stewardship
DuPont Safety, Health & Environment

Cc: Charles Auer – U.S. EPA
Office of Pollution Prevention & Toxics
U. S. Environmental Protection Agency
401 M Street, SW
Washington, DC 20460

Response to EPA's Comments

Physiochemical Properties

EPA Comment (Melting Point): The melting point value provided by the submitter for 2-amino-2-methyl-butanenitrile is not adequate because estimated melting point values above 0°C are not acceptable for the purposes of the HPV Challenge Program. The submitter needs to provide a measured melting point value for this chemical following OECD TG 102. Because the estimated melting point for 2-amino-2-methylpropanenitrile is only slightly below 0°C, and because melting points calculated by EPIWIN often depart from measured values, EPA strongly encourages the submitter to provide measured data for this chemical also.

Response: Melting points for 2-amino-2-methyl-butanenitrile and 2-amino-2-methylpropanenitrile will be measured following OECD Guideline 102. Please note, however, that the purity of available samples of each of the test substances is approximately 75-80%. These materials are prepared in the presence of excess ammonia to stabilize the active substance. The measured melting points will likely be affected by the test substance compositions, and preparation of high-purity test samples may not be technically feasible.

EPA Comment (Boiling Point): The submitter stated that all three chemicals decompose with heat, but did not provide decomposition temperatures. The submitter needs to include this information in the robust summaries.

Response: The test samples will be observed for evidence of decomposition during melting point measurement.

EPA Comment (Vapor Pressure): In the robust summaries, the submitter provided vapor pressure values obtained from company MSDSs. In the test plan the submitter states that these values are measured; the submitter needs to verify this information and include it, with adequate details or citations, in the robust summaries. If these values are estimated, they are not adequate for the purposes of the HPV Challenge Program and the submitter needs to provide measured data for both chemicals following OECT TG 104.

Response: We were unable to locate the original records necessary for responding to the EPA's comments on the studies previously submitted. Therefore, vapor pressure studies following OECD TG 104 with 2-amino-2-methyl-butanenitrile and 2-amino-2-methylpropanenitrile are recommended. However, the commitment to perform these studies is contingent upon their technical feasibility. Specifically, the material currently available is stabilized by addition of excess ammonia and, therefore, may not be suitable for providing a meaningful a vapor pressure measurement. We will determine if it is technically feasible to obtain material of sufficient purity and with the necessary stability to obtain meaningful vapor pressure measurements.

EPA Comment (Water Solubility): The estimated water solubility values provided for both substances are not adequate because estimated water solubility values above 1 μg/L (1 ppb) are not adequate under the HPV Challenge Program. Without more quantitative information about the stability of the chemicals in water, the practicality of testing them for water solubility is unknown. Given the available information, the submitter needs to provide measured water solubility values for the sponsored chemicals following OECD TG 105.

Response: Information to be determined in conjunction with testing of stability in water.

Environmental Fate

EPA Comment (Photodegradation and Fugacity): The submitted data for photodegradation and fugacity are adequate for the purposes of the HPV Challenge Program.

Response: DuPont agrees with this assessment.

EPA Comment (Stability in Water): The submitter needs to provide measured stability in water data, including reaction rates, for both chemicals following OECD TG 111.

Response: We concur that there is a need to perform testing to demonstrate stability in water (e.g., Hydrolysis as a Function of pH, OECD TG 111) for both 2-amino-2-methyl-butanenitrile and 2-amino-2-methylpropanenitrile. TG 111 test is applicable to chemicals that are soluble in water when an analytical technique of sufficient accuracy and precision can be developed. The test will measure the parent chemicals and will also identify the hydrolysis products that occur at a molar concentration of at least 10% of the parent chemical initially added.

EPA Comment (Biodegradation): The submitter needs to provide measured ready biodegradation data for both chemicals following OECD TG 301, taking into account the rate of transformation. If these chemicals react rapidly, then the submitter needs to provide measured ready biodegradation data for the reaction products.

Response: The need for biodegradation testing will be determined based on the results of the TG 111 tests. Substances degraded by water alone would also be expected to degrade at the same rate or faster in biologically active soil in the presence of water. Therefore, we recommend performing the OECD Test Guidance (TG) 301B (CO_2 Evolution Test) to determine biodegradability of the chemicals only if the results of the TG 111 tests at pH 7.0 demonstrate half-lives of the parent chemicals that are greater than 38 days (Jaworska et al., 2003). This half-life is achieved when a chemical is degraded to a level of greater than 40% within 28-days, assuming first-order kinetics.

TG 301B follows the evolution of CO_2 from the biodegradation of the test chemical under aerobic conditions. The test determines if the chemical and its potential metabolites will quickly mineralize to form CO_2 and water. This screening test is

appropriate for test chemicals that are water soluble and have a low vapor pressure (i.e., low Henry's Law Constant), properties that these chemicals are predicted to have. Additionally, the test chemicals should have low or no toxicity to microorganisms at the test concentrations.

Screening tests such as TG 301B are usually first used to determine 'ready biodegradation' when assessing degradation in aqueous systems, (Diderich, 2003). No chemical-specific analyses are usually performed when conducting these tests. This is a stringent test designed to provide limited opportunity for biodegradation and acclimatization of the test chemical. A chemical shown to be ready biodegradable demonstrates that the chemical and its metabolites will rapidly degrade in the environment. Further degradation testing in aqueous systems is not necessary if at least 40% of the Theoretical Carbon Dioxide evolution ($ThCO_2$) is reached within the testing period, as proposed by Jaworska et al. (2003).

A test to determine inherent biodegradation will be considered (e.g., Zahn-Wellens/EMPA Test, OECD TG 302B) to determine if the chemicals will ultimately (bio)degrade in aqueous environments, if they exhibit less than 40% $ThCO_2$ in the CO_2 evolution test and are not toxic at the test concentration. The TG 302B test will determine if the chemicals or their stable metabolites are non-biodegradable/eliminable (<20% biodegradation), partially biodegradable/ eliminable (20 to 70% biodegradation with an indication of the formation of stable metabolites), or inherently biodegradable/eliminable (>70% biodegradation) (Beek, 2001). The proposed testing described above is adequate to characterize the degradation of the sponsored chemicals for the purposes of this program.

References for Environmental Fate

Beek, Bernd, Stella Böhling, Christian Franke, Ulrich Jöhncke, Grabriele Studinger, and Elisabeth Thumm. 2001. Chapter 5, the assessment of biodegradation and persistence. In (ed. By B. Beek) The Handbook of Environmental Chemistry Vol. 2, Part K Biodegradation and Persistence. Springer-Verlag, Berlin. P. 291-320.

Diderich, Robert. 2003. Chapter 8: Environmental Risk Assessment. In (ed. Derek J. Knight and Mike B. Thomas) Practical Guide to Chemical Safety Testing. Rapra Technology Lt. Shawbury, UK p. 163-189.

Jaworska, J.S., R.S. Boethling, and P.H. Howard. 2003. Recent developments in broadly applicable structure-biodegradability relationships. Environ. Tox. Chem. 22(8): 1710-1723.

Health Effects

General Comment

EPA Comment (Closed-System Intermediate): Although the submitter has satisfied many of the criteria for claiming the aminoalkylnitriles as CSI, additional information is

needed to qualify for reduced testing of these chemicals. The submitter has generally addressed the criteria except for the number of sites. The IUR information submitted for the years 1998 and 2002 indicated that the subject chemicals ere manufactured by another company in 2002, and EPA does not have information to document that the chemicals were manufactured and used in closed systems. In addition, there is no information on the disposition of vapors from sampling hoods and from purging of lines used to load the chemicals. The submitter needs to indicate if vapors leaving the laboratory hood are directed to a flare or are otherwise controlled. The submitter also needs to indicate that these chemicals are not present in any products produced using these chemical substances and that there are no releases to the environment in wastes or cleaning solutions. EPA therefore reserved judgment on whether aminoalkylnitriles meet the criteria for CSIs pending the submission of additional information. Conducting a combined repeated dose/ reproductive/developmental toxicity screening test (OECT TG 422) as recommended in the Health Effects section would obviate the need to sustain a CSI claim.

Response: We are unable to address EPA's statement that another company reported manufacture in 2002. Therefore, a repeated dose/developmental toxicity screening test for 2-amino-2-methylpropanenitrile will be performed following OECD Guideline 422.

EPA Comment (Genetic Toxicity – gene mutations): Although the data for the proposed analog in a reverse mutation assay in several strains of *Salmonella typhimurium* with and without metabolic activation appeared adequate in EPA' previous review, the negative results as currently presented do not permit reliable extrapolation to the sponsored chemicals. No information was provided on test substance purity or whether the chemical was cytotoxic. Additional missing study details include incubation conditions (e.g., temperature and duration), criteria for a positive response, mean number of revertant colonies per plate for treated and control cultures, whether or not positive and negative controls gave the appropriate response, and statistical methods. EPA reserves judgement on the adequacy of the gene mutation data pending submission of the missing information. The purity/composition of the tested substance is particularly important in determining whether the data on 2-amino-2,3-dimetylbutanenitrile would adequately represent the toxicity of the sponsored chemicals, given the stated instability of these chemicals.

Response: We have contacted the original submitter of the 2-amino-2,3-dimethylbutanenitrile HPV dossier/test plan. They have agreed to provide the requested information, where available. When the information is received, we will submit a revised dossier/test plan for the aminoalkylnitrile category to the EPA.

EPA Comment (Genetic Toxicity – chromosome aberrations): EPA agrees with the submitter's proposal to conduct an *in vitro* chromosomal aberration test on 2-amino-2-methylpropanenitrile following OECD TG 473.

Response: The *in vitro* chromosomal aberration test following OECD TG 473 will be performed in human lymphocytes.

EPA Comment (Repeated dose and reproductive toxicity): The submitted repeated dose data are inadequate. The 14-day study does not meet the 28-day minimum duration requirements for the repeated dose toxicity endpoint, did not test animals of both sexes, and did not produce any adverse effects at the highest concentration tested. The 28-day study was conducted primarily to examine effects on the nervous system, rather than systemic effects, and the highest tested concentration was much lower then the OECD guideline requires. Without information on the relative rates of disproportionation in water and the importance of the reaction products to the overall toxicity of the category, it is unclear whether the data generated on 2-amino-2,3-dimethylbutanenitrile can represent the toxicity of the category. Testing is needed to address repeated dose toxicity.

Response: A repeated dose/reproductive/developmental toxicity screening test for 2-amino-2-methylpropanenitrile will be performed following OECD Guideline 422.

Ecological Effects

EPA Comment (Fish): Missing study details include hardness and total organic carbon of the dilution water, temperature, number of fish per test vessel, fish loading, mean fish length/weight, and 95% confidence limits.

Response: Where available, data was added to address the missing/requested information. A revised robust summary is included with this submission.

EPA Comment (Invertebrates): The robust summary for *Daphnia* 48-hour static toxicity of 2-amino-2-methylpropanenitrile is missing the following details: hardness and total organic carbon of the dilution water, temperature, pH, dissolved oxygen, age of the daphnids, and 95% confidence limits. The robust summary for *Daphina* 48-hour static toxicity of 2-amino-2,3-dimethylbutane is missing test substance purity, hardness and total organic carbon of the dilution water, and age of the daphnids. Dissolved oxygen and pH values were measured during the study, but were not reported in the summary.

Response: Where available, data was added to address the missing/requested information for the study.

EPA Comment (Algae): Missing study details for 2-amino-2,3-dimethylbutanenitrile include test substance purity, lighting conditions, pH, and whether the EC_{50} was based on measured or nominal concentrations. References to the statistical determination of an LC_{50} should be revised to reflect determination of an EC_{50}.

Response: Where available, data was added to address the missing/requested information. A revised robust summary is included with this submission.